Plant Cell, Tissue and Organ Culture (PCTOC)
https://doi.org/10.1007/s11240-022-02252-1

ORIGINAL ARTICLE

Enhancement of phenolic acids accumulation in *Salvia abrotanoides* (Kar.) Sytsma shoot cultures under elicitation with nitric oxide

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Received: 13 November 2021 / Accepted: 9 February 2022

Abstract

Salvia abrotanoides (Kar.) Sytsma is a medicinal plant that contains phenolic acids, especially rosmarinic acid (RA), with a wide variety of health benefits. In this research, the stimulatory effects of sodium nitroprusside (SNP), a donor of nitric oxide (NO), were examined on phenolic acids accumulation in the shoot cultures of the species after different times of exposure. The developed shoots on single-nodal explants in Murashige and Skoog solid medium supplemented with 0.5 mg L^{-1} kinetin and 0.05 mg L^{-1} indole-3-acetic acid were treated with SNP at concentrations of 25, 50 and 100 µM. The highest content of RA (10.45 ± 0.40 mg g^{-1} DW) was measured 144 h after elicitation of the shoots with 100 µM SNP. The maximum values of salvianolic acid A (Sal-A) (0.065 ± 0.00 mg g^{-1} DW) and salvianolic acid B (Sal-B) (0.42 ± 0.01 mg g^{-1} DW) were obtained in the shoots after 96 and 144 h exposure to 50 and 25 µM SNP, respectively. Also, elicitation with SNP at different concentrations significantly upregulated the crucial genes (*PAL*, *TAT*, *RAS* and *CYP98A14*) involved in phenolic acids biosynthesis in the shoots with distinct patterns, although no strong correlations were observed between transcription levels of the genes and phenolic acids accumulation. The findings of this study provide beneficial information about the impact of NO as an effective elicitor, which could be valuable for the in vitro improvement of phenolic acids production in *S. abrotanoides*.

Key Message

The results demonstrated that the elicitation with nitric oxide upregulated critical genes in the biosynthetic pathways and led to phenolic acids accumulation in the shoots of *Salvia abrotanoides* (Kar.) Sytsma.

Keywords *Salvia abrotanoides* · Nitric oxide · Phenolic acids · Gene expression · Shoot culture

Abbreviations

KIN	Kinetin
IAA	Indole-3-acetic acid
FSI	Frequency of shoot initiation
MSN	Mean shoot number
MSL	Mean shoot length
NO	Nitric oxide
SNP	Sodium nitroprusside
TPC	Total phenolic content
TFC	Total flavonoid content
RA	Rosmarinic acid
Sal-A	Salvianolic acid A
Sal-B	Salvianolic acid B
PAL	Phenylalanine ammonia-lyase
TAT	Tyrosine aminotransferase
RAS	Rosmarinic acid synthase
CYP98A14	Cytochrome P450-dependent monooxygenase

Introduction

Medicinal plants are the primary sources of valuable bioactive compounds principally used in the pharmaceutical, cosmetic and food industries (Lubbe and Verpoorte 2011). Such biologically active compounds are abundant in many plants from the Lamiaceae family (Trivellini et al. 2016). *Salvia* L. is the largest and most widespread genus of the mint family that grows abundantly in the Mediterranean

Communicated by Patricia Marconi.

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Published online: 08 March 2022